

What is claimed is:

1 1. A method comprising:

2 (a) obtaining a nucleic acid molecule comprising a
3 chimeric gene, said chimeric gene comprising a first portion
4 and a second portion, the first portion encoding a *KRE9*
5 lacking a functional signal sequence and the second portion
6 being a heterologous nucleic acid sequence;

7 (b) transforming a yeast cell lacking a functional
8 *KRE9* gene with said nucleic acid molecule; and

9 (c) determining whether said transformed yeast cell
10 grows when supplied with a medium that permits growth of a
11 yeast cell expressing *KRE9* having a functional signal
12 sequence, but does not permit growth of a yeast cell that
13 does not express *KRE9* having a functional signal sequence,
14 wherein growth on said medium indicates that said
15 heterologous nucleic acid sequence present in said yeast
16 cell encodes a signal sequence.

1 2. The method of claim 1, wherein step (a)
2 comprises:

3 (i) obtaining double-stranded DNA;

4 (ii) ligating said double-stranded DNA to a DNA
5 molecule encoding *KRE9* lacking a functional signal sequence
6 to create a chimeric gene.

1 3. The method of claim 1, wherein step (a)
2 comprises:
3 (i) obtaining double-stranded DNA;
4 (ii) ligating said double-stranded DNA to a DNA
5 molecule encoding *KRE9* lacking a functional signal sequence
6 to create a chimeric gene;
7 (iii) transforming a bacterium with said nucleic
8 acid molecule comprising a chimeric gene;
9 (iv) growing said transformed bacterium; and
10 (v) isolating said nucleic acid molecule comprising
11 a chimeric gene from said transformed bacterium.

1 4. The method of claim 1, further comprising, in
2 order to identify said signal sequence, isolating and
3 sequencing a portion of the chimeric gene contained within a
4 yeast cell that grows when supplied with a medium that
5 permits growth of a yeast cell expressing *KRE9*, but does not
6 permit growth of a yeast cell that does not express *KRE9*
7 having a functional signal sequence.

1 5. The method of claim 1, wherein said second
2 portion of said nucleic acid molecule is pBOSS1.

1 6. The method of claim 1, wherein said second
2 portion of said nucleic acid molecule is cDNA.

1 7. The method of claim 1, wherein the yeast strain
2 is Yscreen2.

1 8. The method of claim 1, wherein said medium
2 contains glucose as the sole carbon source.

1 9. The method of claim 8, wherein the medium
2 contains a calcineurin inhibitor.

1 10. The method of claim 4, further comprising using
2 a nucleic acid molecule encoding said signal sequence to
3 screen an eukaryotic library for a full-length gene or cDNA
4 encoding a protein comprising said identified signal
5 sequence.

1 11. A yeast cell transformed with a nucleic acid
2 molecule comprising a chimeric gene, said chimeric gene
3 comprising a first portion and a second portion, the first
4 portion encoding a *KRE9* lacking a functional signal sequence
5 and the second portion being a heterologous nucleic acid
6 sequence.

1 12. A method comprising:

2 (a) obtaining a nucleic acid molecule comprising a
3 chimeric gene, said chimeric gene comprising a first portion
4 and a second portion, the first portion encoding a *KRE9*
5 lacking a functional signal sequence and the second portion
6 being a heterologous nucleic acid sequence;

7 (b) transforming a yeast cell lacking a functional
8 *KRE9* gene with said nucleic acid molecule; and

9 (c) determining whether said transformed yeast cell
10 grows when supplied with a medium that does not permit
11 growth of a yeast cell expressing *KRE9* having a functional
12 signal sequence, but does permit growth of a yeast cell that
13 does not express *KRE9* having a functional signal sequence,
14 wherein lack of growth on said medium indicates that said
15 heterologous nucleic acid sequence present in said yeast
16 cell encodes a signal sequence.

1 13. The method of claim 12, wherein the medium
2 contains K1 killer toxin.

1 14. The method of claim 12, wherein step (a)
2 comprises:
3 (i) obtaining a double-stranded DNA; and
4 (ii) ligating said double-stranded DNA to a DNA
5 molecule encoding *KRE9* lacking a functional signal sequence
6 to create a chimeric gene.

1 15. The method of claim 12, further comprising, in
2 order to identify said signal sequence, isolating and
3 sequencing a portion of the chimeric gene contained within
4 said yeast cell that does not grow when supplied with a
5 medium that does not permit growth of a yeast cell
6 expressing *KRE9*, but does permit growth of a yeast cell that
7 does not express *KRE9* having a functional signal sequence.

1 16. The expression vector pBOSS-1.

1 17. A genetically engineered host cell comprising
2 the vector of claim 16.